

# **Development and Field Application of Laser Particle Imagers**

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## **LONG-TERM GOAL**

The long term goal is development and utilization of a suite of laser based particle imaging systems as part of a comprehensive marine particle analysis system. The system is designed with wide dynamic range, thus, it will ultimately be used for high speed, high resolution characterization of water column particle fields in high, medium and low latitudes. Such a system will have broad application to areas of defense and environmental interests such as ecological modeling efforts (e.g. HYCODE/ECOHAB), ASON, and satellite calibration.

## **OBJECTIVES**

The project has two major objectives:

- 1) Continuation of technical development of our particle sensors (SIPPER) and (MIPPER) in areas such as image resolution and quality, ease of use, durability and integration with other platforms
- 2) Application of those sensors to field programs supporting defense/environmental biophysical models and zooplankton spatial distribution models. The latter is a critical component governing planktic interactions which previous methods (less fine-scale) have not been able to examine adequately.

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## APPROACH

The approach for this project is to improve upon the previously developed sensor (SIPPER) while developing a new sensor (MIPPER) designed to examine the distribution of smaller forms of plankton that are missed by SIPPER. Both sensors are being flume tested and calibrated at the USF Center for Ocean Technology (COT) and sea-tested as part of our high resolution sampler (HRS) towed platform. These sea-tests also serve as the field application of the sensors by collecting information for the HYCODE/ ECOHAB modeling of zooplankton grazing and our new initiative of the examination of zooplankton microdistribution on a diel basis.

## WORK COMPLETED

### *Sensor Development: Shadowed Image Particle Profiling and Evaluation Recorder (SIPPER) and Miniature Image Particle Profiling and Evaluation Recorder (MIPPER)*

The SIPPER instrument has been developed to accurately evaluate particle concentrations in a wide range of sizes, by providing as its output high-quality digital images. This instrument is uniquely capable of completely imaging particles from several tens of microns through several centimeters in length, from two directions [1].

Several improvements to the SIPPER instrument have been designed, realized, and tested at sea. These are being incorporated into MIPPER. Changes have been made to make the system simpler to use. Control of the system is done over a TCP/IP Ethernet link. A low power chip-embedded web server (*SitePlayer<sup>TM</sup>*) contains a custom web page and digital I/O pins, and allows the user to control SIPPER via a simple web-page interface. We have added an SDSL link (1.168Mbps over 3352 meters, 272kbps over 5182 meters) to relay Ethernet command information to the underwater instrument, over a single twisted pair in the 500m long electromechanical tether between the ship and HRS. The SDSL transceiver, along with a 10/100 Base-T Ethernet switch, 10- or 100-FT fiber-Ethernet converters, and an RS-232 to Ethernet converter, are housed in a new 150 mm diameter pressure vessel, and attached within the HRS frame. The optical fiber-Ethernet and RS232 links are included to enable future upgrades to HRS instrumentation.

Two higher resolution cameras (*Dalsa Corporation*), capable of imaging at 86,000 Mpixels per second each have been integrated into the existing SIPPER imaging pressure vessels. The camera sensors have a slightly larger imaging width compared to the EG&G cameras used previously, necessitating changes to the optical component positions. Increasing resolution of the camera resulted in sharp images, but only at the focal plane. An optical f-stop has been added to allow us to balance depth of field and resolving power over the ~100 cm<sup>2</sup> imaging cross-section.

A real-time lighting correction and data compression scheme has been implemented using *Xilinx* FPGAs. These additions improve image quality and simplify data storage and post-processing requirements. They also help to significantly (~200x) reduce the data rate and enable quick changes to the system. The data storage system for SIPPER has been upgraded from a standalone 50GB SCSI-disk based system (7.5MBps) requiring UNIX-based data offload to a new system (Boulder Instruments) that enables direct and continuous streaming of data from the new data processing board to three Ultra-ATA drives (120GB total) at up to 21MBps. The new storage system allows the user to quickly and easily offload to a network drive over a 100-BaseT-network link that is attached once the

HRS is on deck. The increased data capacity and compression has allowed grayscale images of particles to be stored, even in particle-rich waters. We have developed an adaptive threshold that computes 3-bit grayscale from the original 8-bit images. The threshold is based on the lighting correction, so maximum use of the three bits is assured. Real-time compression of grayscale data is again accomplished in the FPGA. A virtual switch on the web page allows the user to immediately change from black and white to gray scale imaging without opening any pressure vessels or bringing the instrument shipboard. In-tube flow rate is available so tow speed can be adjusted as desired.

Software has been optimized to enable decompression and particle image extraction in a timely manner (approximately 20 minutes for 10 minutes of image collection, versus 20 hours previously). Software has also been created to decompress and generate frame images from acquired grayscale data. These images are being made available to another ONR project (N00014-02-1-0266), for development of automated plankton image analysis software.

In summary, improvements to the SIPPER electronics and software during the last year have resulted in:

- A better user interface in real-time
- Faster downloading of the data
- Faster decompression of files (by two orders of magnitude) and extraction of particle image bitmaps
- Increased image resolution and gray scale output which in turn improves image identification

MIPPER is being developed as a reduced-size and imaging volume ( $9\text{ cm}^2$  vs  $100\text{ cm}^2$ ) version of SIPPER. Its aim is to image the smaller size fraction (50-500  $\mu\text{m}$  plankton) that is imaged by SIPPER, but with higher resolution, which will enhance identification of this biologically important size range. Because of outside interest in the SIPPER instrument, especially in moored or buoyed applications, MIPPER is being developed to operate as a pump-assisted instrument in addition to its open flow mode. Much of the lessons learned in the development of SIPPER are directly applicable to the MIPPER instrument.

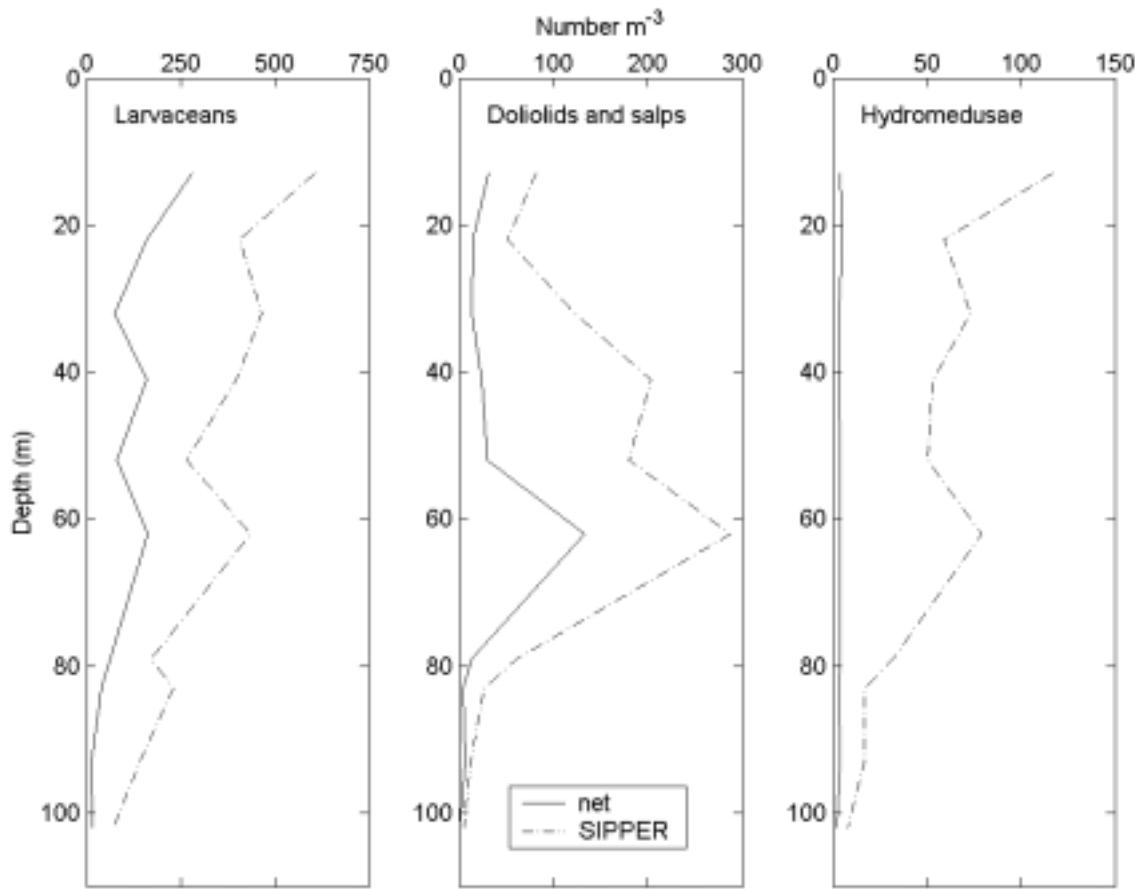
#### *Field Application*

During the last year two cruises were conducted on the West Florida Shelf (WFS) with SIPPER deployed on the HRS. On a cruise in November 2001 the dual camera system on SIPPER was tested and data was collected for the seasonal survey of zooplankton grazing in the HYCODE/ECOHAB test site. This completes our seasonal survey and a manuscript is currently being prepared. A second cruise was completed in September 2002 testing the SIPPER with the new gray scale capabilities and involving sampling over a twenty-four period for the examination of diel microdistribution patterns.

## **RESULTS**

Analysis of a SIPPER dataset collected in the summer of 2000 has been completed. That study compared the abundance and size distribution of mesozooplankton collected by the HRS plankton net carousel and SIPPER imagery collected concurrently inside the sampling duct leading to the net carousel in offshore waters of the Gulf of Mexico [2]. Approximately 175,000 SIPPER bitmaps were manually examined and classified from 10 depths fished between 10 and 100 meters. We found that the nets significantly underestimated both abundance (35% of SIPPER total) and biomass (46 % of

SIPPER total) of the mesozooplankton when matched against the SIPPER imagery. Much of the discrepancy was due to the limitations of nets in efficiently sampling the fragile and gelatinous components of the plankton, as they tend to destroy or extrude these forms. For example, larvaceans (300%), doliolids and salps (380%) and hydromedusae (1400%) were all significantly more abundant when sampled optically (fig 1). Biomass was also much higher for these groups in the SIPPER data when compared against the nets. These differences are so great that the composition and trophodynamic structure of the mesozooplankton assemblage are radically different than would be determined using traditional methods. For instance, our group has found that larvaceans are a primary

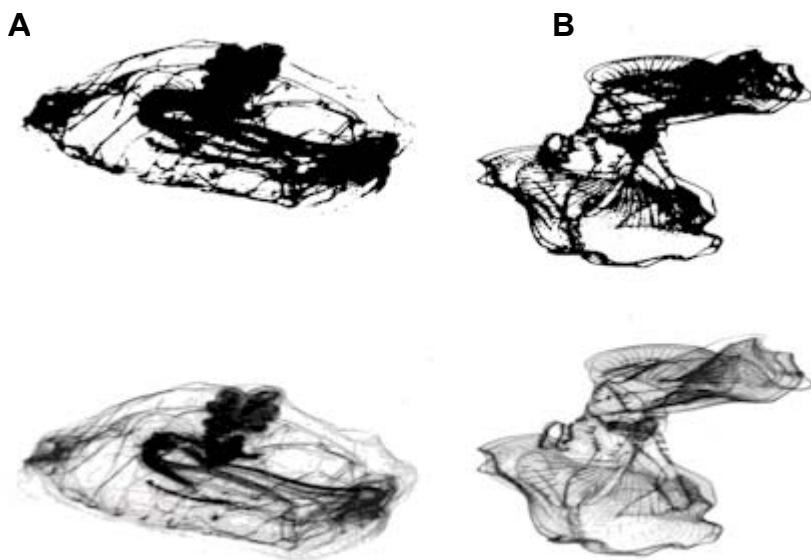


**Figure 1 Net and SIPPER abundance comparison of three soft-bodied zooplankton groups (Larvaceans, doliolids and hydromedusae)**

grazer on the WFS, site of a HyCODE-ECOHAB modeling site, at times accounting for 66% of the grazing pressure [3]. If the sampling biases identified from this current study hold true, larvaceans would be an even more significant source of phytoplankton loss in this study area.

Our cruise in November 2001 represents the completion of the seasonal survey of zooplankton in the HYCODE/ECOHAB study area. It will be combined with four other cruises to provide data on seasonal grazing to the modeling effort of HyCODE-ECOHAB. This cruise was also the first in which a *Karenia brevis* red tide bloom was sampled by the HRS. We were thus able to examine the spatial distribution of zooplankton in relation to the bloom with the HRS (10 nets towed at different depths) and SIPPER system. We found several zooplankton groups that appeared to avoid the bloom. This would be a unique observation and a potentially critical one for how grazing may affect these harmful alga blooms.

The cruise just completed in September 2002 was designed to examine the diel distribution patterns of mesozooplankton at the HyCODE-ECOHAB site and an oceanic sampling station. The cruise was cut short by the presence of a hurricane in the Gulf, but one twenty-four series was collected and will be examined over the coming months. This cruise also represented our first deployment of SIPPER enabled for gray scale imaging, which proved a tremendous improvement. Many zooplankton groups exhibit varying degrees of transparency and the addition of gray scale allows these to be visualized (fig. 2). The improved image quality makes image identification much easier and gray scale imagery will be incorporated into the automated pattern recognition being developed under grant ONR N00014-02-1-0266. Gray scale images were made possible by improvements to the SIPPER electronics and data storage system and should pay major ecological dividends in applications.



**Figure 2. Examples of SIPPER imaged organisms (A. Salp, B. pseudothecosome pteropod) grayscale (bottom images) and after binary threshold had been implemented (top images) demonstrating improved image quality using grayscale.**

## **IMPACT/APPLICATIONS**

This project represents a directed effort to develop, build and deploy laser-based imaging systems that can characterize the oceanic particle spectrum in sizes from centimeters to micrometers and be deployed on AUVs, ROVs and towed or moored sampling systems. As such it is a part of the larger goals of much ONR-directed research to create systems that can efficiently monitor oceanic conditions with a minimum of temporal and spatial degradation and without the use of expensive and time-consuming research vehicles. Such information would then become a major input to forecasting (or ‘nowcasting’) significant conditions in the coastal ocean.

## **TRANSITIONS**

The data collected with SIPPER and MIPPER are being provided to the HYCODE/ECOHAB modeling group (Walsh and Weisberg, ONR #N00014-99-1-0212) to set variable ranges for zooplankton grazing. The instruments are also being used to examine the specific distribution and abundance of the cyanobacteria *Trichodesmium*. *Trichodesmium* is able to fix nitrogen and has been identified as a major contributor to nitrogen flux in the world ocean [4]. On the West Florida Shelf it may be a necessary precursor to red tides [5]. Our data will be provided to other researchers at USF with separately funded proposals examining these relationships.

## **RELATED PROJECTS**

Development of software enabling automated classification of SIPPER images is being funded through ONR award # N00014-02-1-0266. The SIPPER web site is at <http://marine.usf.edu/sipper>.

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2. Remsen, A., Samson, S., and Hopkins, T. “What you see is not always what you get: Comparison of a Zooplankton-Imaging Sensor (SIPPER) With Concurrent Optical Plankton Counter and net Data From the Gulf of Mexico”, AGU-ASLO Ocean Sciences Meeting, Talk OS-41M-09, Honolulu, HI, Feb. 11-17, 2002.
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2. Remsen, A., Samson, S., and Hopkins, T. "What you see is not always what you get: Comparison of a Zooplankton-Imaging Sensor (SIPPER) With Concurrent Optical Plankton Counter and net Data From the Gulf of Mexico", AGU-ASLO Ocean Sciences Meeting, Talk OS-41M-09, Honolulu, HI, Feb. 11-17, 2002.